ASSESSING THE CYTOTOXICITY OF BLACK CARBON AS A MODEL FOR ULTRAFINE ANTHROPOGENIC AEROSOL ON HUMAN EPITHELIAL LUNG CELLS AND MURINE MACROPHAGES

M. Esther Salinas, M.S.,[1,2] Kristine M. Garza, Ph.D.,[1,2] Thomas E. Gill, Ph.D., P.G.[1,3]

1 Environmental Science and Engineering Program, University of Texas- El Paso, El Paso, TX, USA
2 Department of Biological Sciences, University of Texas- El Paso, El Paso, TX, USA
3 Department of Geological Sciences, University of Texas- El Paso, El Paso, TX, USA

Combustion-derived nanomaterials or ultrafine (<1 μm) atmospheric aerosols are primarily products of anthropogenic activities, such as the burning of fossil fuels. Ultrafine particles (UFPs) can absorb other noxious pollutants including volatile organic compounds (VOCs), polycyclic aromatic hydrocarbons (PAHs), toxic organic compounds, and heavy metals. The combination of high population density, meteorological conditions, and industrial productivity brings high levels of air pollution to the metropolitan area of El Paso, Texas, USA/ Ciudad Juarez, Chihuahua, Mexico, comprising the Paso del Norte air basin. A study conducted at Research Triangle Park, NC analyzed sites adjacent to heavy-traffic highways in El Paso and elucidated higher UFP concentrations in comparison to previously published work exploring pollution and adverse health effects in the basin. UFPs can penetrate deep into the alveolar sacs of the lung, reaching distant alveolar sacs and inducing a series of immune responses that are detrimental to the body: evidence suggests that UFPs can also cross the alveolar-blood barrier and potentially endanger the body's immune response. The properties of UFPs and the dynamics of local atmospheric and topographical conditions indicate that emissions of nanosized carbonaceous aerosols could pose significant threats to biological tissues upon inhalation by residents of the Paso del Norte.

This study utilizes Black Carbon (BC) as a model for environmental UFPs and its effects on the immunological response. An *in vitro* approach is used to measure the ability of BC to promote cell death upon long-term exposure due to its feasibility, reproducibility, and low-cost of overall laboratory needs. Human epithelial lung cells (A549) and murine macrophages (RAW264.7) were treated with BC and assessed for metabolic activity after chronic exposure. The cell viability experiments included a chronic study at 7, 10, and 14 days of UFP exposure at six different concentrations of BC: 1x, 3x, 6x, 12.5x, 25x, and 50x conducting the following assays: Trypan blue exclusion test, Calcein-AM viability assay, and CellTiter- Glo viability assay. The trypan blue exclusion test determines the number of viable cells in a suspension based on the principle that live cells have membranes that remain intact and exclude dyes like trypan blue, whereas dead cells do not contain that intact barrier. Calcein-AM is a cell-permeant dye that undergoes hydrolysis by live cells via mitochondrial enzymes, yielding a fluorescent product. CellTiter- Glo luminescent assay calculates metabolically active cell proliferation by quantitating the ATP concentration present in the culture. Graphical data and numerical results on the various cytotoxicity analyses will be displayed in a Poster Presentation for the International Conference on Carbonaceous Particles in the Atmosphere, 2015 at the Lawrence Berkeley National Lab in Berkeley, CA.

The research conducted will help identify constituents of the Paso del Norte aerosol that specifically contribute to increased incidence of respiratory ailments and to help strategically develop therapies for local residents adversely impacted by these aerosols. Follow up research will investigate how chronic exposure to BC induces oxidative stress and activates cytokine production, processes concomitant with diseases of the lung, and ultimately improve understanding of the mechanisms of impact of nanosized PM on human health.